

**What is claimed is:**

1. A method of bioconversion using a biocatalyst, which comprises the steps of:

- (a) preparing a vector for spore surface display comprising a gene construct containing a gene encoding a display motif and a gene encoding the biocatalyst, wherein, when expressed, the gene construct expresses the display motif and the biocatalyst in a fusion form and the biocatalyst is displayed on a spore surface;
- 5 (b) transforming a host cell with the vector for spore surface display;
- 10 (c) displaying the biocatalyst on the spore surface of the host cell;
- 15 (d) recovering the spore displaying on its surface the biocatalyst; and
- (e) performing the bioconversion reaction using the spore displaying on its surface the biocatalyst.

2. A method of bioconversion using a biocatalyst, which comprises the steps of:

- (a) transforming a host cell harboring a genetic carrier selected from the group consisting of spore and virus with a vector containing a gene encoding the biocatalyst;
- 25 (b) culturing the transformed host cell and expressing the biocatalyst in the host cell;
- (c) allowing to form noncovalent bonds between the

expressed biocatalyst and a surface of the genetic carrier so that the biocatalyst is displayed on the surface of the genetic carrier;

5 (d) recovering the genetic carrier displaying on its surface the biocatalyst; and

(e) performing the bioconversion reaction using the genetic carrier displaying on its surface the biocatalyst.

10 3. The method according to claim 1 or 2, wherein the spore is derived from a spore-forming Gram negative bacterium including *Myxococcus*, a spore-forming Gram positive bacterium including *Bacillus*, a spore-forming *Actionmycete*, a spore-forming yeast or a spore-forming  
15 fungus.

4. The method according to claim 3, wherein the spore is derived from a spore-forming Gram positive bacterium.

20 5. The method according to claim 4, wherein the spore is derived from *Bacillus*.

6. The method according to claim 1, wherein the display motif is derived from a spore coat protein.

7. The method according to claim 6, wherein the spore coat protein is selected from the group consisting of Cota,

CotB, CotC, CotD, CotE, CotF, CotG, CotH, CotJA, CotJC,  
CotK, CotL, CotM, CotS, CotT, CotV, CotW, CotX, CotY, CotZ,  
SpoIVA, SspoVID and SodA.

5 8. The method according to claim 6, wherein the spore  
coat protein is a modified form of one selected from the  
group consisting of CotA, CotB, CotC, CotD, CotE, CotF,  
CotG, CotH, CotJA, CotJC, CotK, CotL, CotM, CotS, CotT,  
CotV, CotW, CotX, CotY, CotZ, SpoIVA, SspoVID and SodA, in  
10 which the modified form has a more compatibility for spore  
surface display relative to wild type spore coat protein.

9. The method according to claims 8, wherein the  
modification of the spore coat protein is obtained by  
15 mutating a gene encoding the spore coat protein according  
to a method selected from the group consisting of DNA  
shuffling method, STEP method, RPR method, molecular  
breeding method, ITCHY method, error-prone PCR, point  
mutagenesis, nucleotide mutagenesis, combinatorial  
20 cassette mutagenesis and other suitable random mutagenesis.

10. The method according to claim 7 or 8, wherein the  
spore coat protein is CotE or CotG.

25 11. The method according to claim 1, wherein the surface  
motif is derived from randomly-synthesized peptides.

12. The method according to claim 1, wherein the surface motif is a peptide or polypeptide selected from a natural- occurring random library.

5 13. The method according to claim 1 or 2, wherein the biocatalyst is selected from the group consisting of a hydrolase, an oxidoreductase, a transferase, a lyase, an isomerase and a ligase.

10 14. The method according to claim 13, wherein the biocatalyst is a transferase.

15. The method according to claim 14, wherein the transferase is an enzyme catalyzing transglycosylation.

15  
16. The method according to claim 15, wherein the enzyme catalyzing transglycosylation is  $\beta$ -galactosidase, levansucrase, dextranucrase, inulosucrase, glycogen synthase, chitin synthase, starch synthase,  
20 cyclomaltodextrin glucanotransferase or 4- $\alpha$ -glucanotransferase.

17. The method according to claim 1, wherein the fusion form of the display motif and the biocatalyst has an order 25 of the display motif-the biocatalyst or the biocatalyst- the display motif.

18. The method according to claim 1 or 2, wherein the biocatalysts displayed on spore surface are covalently crosslinked.

5 19. The method according to claim 1 or 2, wherein the biocatalyst exhibits one or more stability selected from the group consisting of thermal stability, pH stability, a resistance to organic solvent, stability to high-concentrated salt and stability to dry, in which the  
10 stability of the biocatalyst is enhanced compared to a free biocatalyst.

20. The method according to claim 1 or 2, wherein the spore exhibits lower protease activity or no protease  
15 activity.

21. The method according to claim 1 or 2, wherein the spore is non-reproductive one.

20 22. The method according to claim 2, wherein the virus is a bacteriophage.

23. The method according to claim 2, wherein the biocatalyst is modified one by virtue of: (i) deleting a  
25 portion of amino acids of the biocatalyst; (ii) fusing oligopeptide or polypeptide, which enhances noncovalent bond between the biocatalyst and a surface protein of the

spore or virus, to the biocatalyst; (iii) subjecting the biocatalyst to site-directed mutagenesis; or (iv) subjecting the biocatalyst to random mutagenesis.

5 24. The method according to claim 23, wherein the biocatalyst modified by deleting a portion of amino acids is prepared by deleting ionic amino acids from N-terminal sequence of the biocatalyst.

10 25. The method according to claim 23, wherein the biocatalyst modified is prepared by fusing cationic peptide to the biocatalyst.

26. The method according to claim 2, wherein the spore or 15 virus is modified by virtue of: (i) fusing oligopeptide or polypeptide, which enhances noncovalent bond between the biocatalyst and a surface protein of the spore or virus, to its surface protein; (ii) subjecting the surface protein to site-directed mutagenesis; or (iii) subjecting 20 the surface protein to random mutagenesis.

27. The method according to claim 2, wherein the biocatalyst has covalent bonds (i) between spore or virus surface and the biocatalyst; or (ii) between the 25 biocatalysts.

28. The method according to claim 27, wherein the covalent

bond is formed by a chemical method including glutaraldehyde treatment, a physical method including ultraviolet treatment, or a biochemical method including enzyme treatment to allow the formation of covalent bond.

5

29. A biocatalyst displayed on a spore surface and fused covalently to a display motif.

30. A biocatalyst displayed on a spore or virus surface by  
10 virtue of noncovalent bonds.

31. The biocatalyst according to claim 29 or 30, wherein  
the spore is derived from a spore-forming Gram negative  
bacterium including *Myxococcus*, a spore-forming Gram  
15 positive bacterium including *Bacillus*, a spore-forming  
*Actionmycete*, a spore-forming yeast or a spore-forming  
fungus.

32. The biocatalyst according to claim 31, wherein the  
20 spore is derived from a spore-forming Gram positive  
bacterium.

33. The biocatalyst according to claim 32, wherein the  
spore is derived from *Bacillus*.

25

34. The biocatalyst according to claim 29, wherein the  
display motif is derived from a spore coat protein.

35. The biocatalyst according to claim 34, wherein the  
spore coat protein is selected from the group consisting  
of CotA, CotB, CotC, CotD, CotE, CotF, CotG, CotH, CotJA,  
5 CotJC, CotK, CotL, CotM, CotS, CotT, CotV, CotW, CotX,  
CotY, CotZ, SpoIVA, SspoVID and SodA.

36. The biocatalyst according to claim 34, wherein the  
spore coat protein is a modified form of one selected from  
10 the group consisting of CotA, CotB, CotC, CotD, CotE, CotF,  
CotG, CotH, CotJA, CotJC, CotK, CotL, CotM, CotS, CotT,  
CotV, CotW, CotX, CotY, CotZ, SpoIVA, SspoVID and SodA, in  
which the modified form has a more compatibility for spore  
surface display relative to wild type spore coat protein.

15

37. The biocatalyst according to claims 36, wherein the  
modification of the spore coat protein is obtained by  
mutating a gene encoding the spore coat protein according  
to a method selected from the group consisting of DNA  
20 shuffling method, STEP method, RPR method, molecular  
breeding method, ITCHY method, error-prone PCR, point  
mutagenesis, nucleotide mutagenesis, combinatorial  
cassette mutagenesis and other suitable random mutagenesis.

25 38. The biocatalyst according to claim 35 or 36, wherein  
the spore coat protein is CotE or CotG.

39. The biocatalyst according to claim 29, wherein the surface motif is derived from randomly-synthesized peptides.

5 40. The biocatalyst according to claim 29, wherein the surface motif is a peptide or polypeptide selected from a natural-occurring random library.

10 41. The biocatalyst according to claim 29 or 30, wherein the biocatalyst is selected from the group consisting of a hydrolase, an oxidoreductase, a transferase, a lyase, an isomerase and a ligase.

15 42. The biocatalyst according to claim 41, wherein the biocatalyst is a transferase.

43. The biocatalyst according to claim 42, wherein the transferase is an enzyme catalyzing transglycosylation.

20 44. The biocatalyst according to claim 43, wherein the enzyme catalyzing transglycosylation is  $\beta$ -galactosidase, levansucrase, dextranucrase, inulosucrase, glycogen synthase, chitin synthetase, starch synthase, cyclomaltodextrin glucanotransferase or 4-a-glucanotransferase.

45. The biocatalyst according to claim 29, wherein the

fusion form of the display motif and the biocatalyst has an order of the display motif-the biocatalyst or the biocatalyst-the display motif.

5 46. The biocatalyst according to claim 29 or 30, wherein the biocatalysts displayed on spore surface are covalently crosslinked.

10 47. The biocatalyst according to claim 29 or 30, wherein the biocatalyst exhibits one or more stability selected from the group consisting of thermal stability, pH stability, a resistance to organic solvent, stability to high-concentrated salt and stability to dry, in which the stability of the biocatalyst is enhanced compared to a 15 free biocatalyst.

48. The biocatalyst according to claim 29 or 30, wherein the spore exhibits lower protease activity or no protease activity.

20 49. The biocatalyst according to claim 29 or 30, wherein the spore is non-reproductive one.

50. The biocatalyst according to claim 30, wherein the 25 virus is a bacteriophage.

51. The biocatalyst according to claim 30, wherein the

biocatalyst is modified one by virtue of: (i) deleting a portion of amino acids of the biocatalyst; (ii) fusing oligopeptide or polypeptide, which enhances noncovalent bond between the biocatalyst and a surface protein of the 5 spore or virus, to the biocatalyst; (iii) subjecting the biocatalyst to site-directed mutagenesis; or (iv) subjecting the biocatalyst to random mutagenesis.

52. The biocatalyst according to claim 51, wherein the 10 biocatalyst modified by deleting a portion of amino acids is prepared by deleting ionic amino acids from N-terminal sequence of the biocatalyst.

53. The biocatalyst according to claim 51, wherein the 15 biocatalyst modified is prepared by fusing cationic peptide to the biocatalyst.

54. The biocatalyst according to claim 30, wherein the spore or virus is modified by virtue of: (i) fusing 20 oligopeptide or polypeptide, which enhances noncovalent bond between the biocatalyst and a surface protein of the spore or virus, to its surface protein; (ii) subjecting the surface protein to site-directed mutagenesis; or (iii) subjecting the surface protein to random mutagenesis.

55. The biocatalyst according to claim 30, wherein the biocatalyst has covalent bonds (i) between spore or virus

surface and the biocatalyst; or (ii) between the biocatalysts.

56. The biocatalyst according to claim 55, wherein the  
5 covalent bond is formed by a chemical method including  
glutaraldehyde treatment, a physical method including  
ultraviolet treatment, or a biochemical method including  
enzyme treatment to allow the formation of covalent bond.